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**Original article**

# Liquefying crystallized honey with ultrasonic waves

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**Summary** — Ten samples of crystallized honey were divided into 3 equal parts. The first was liquified by ultrasonic waves at 23 kHz, the second by heating at 60°C for 30 mins and the third remained untreated. The time that was needed for the liquefaction of honey varied from 18 to 25 min, the energy from 0.1056 to 0.1466 kWh, and the temperature from 76 to 82°C. The quality of honey was affected by increasing hydroxymethylfurfural (HMF) levels from 6.5 ± 0.8 ppm to 11.4 ± 1.4 ppm and decreasing diastase activity from 20.8 ± 1.8 DN to 17.4 ± 1.5 DN. Significant differences ($p < 0.010$) were found among the HMF averages of samples liquefied by sonication and samples liquefied by heating. No differences were found among diastase activity, moisture, electrical conductivity, pH and time of recrystallization among ultrasonic and heat treatment.

**crystallisation / honey / ultrasound / HMF / diastase**

## INTRODUCTION

Ultrasonic waves are sound waves with a higher frequency than the maximum that can be sensed by the human ear. These waves are transmitted through water without being weakened in power. They cause mechanical and thermic changes in the material through which they pass, and cause changes in unicellular organisms; they may have many other effects. Due to their attributes they are increasingly used in modern technology, science and medicine.

In honey technology, Kalogereas (1955), reported that high frequency sound waves (9 kHz) eliminated the existing crystals and retarded further crystallization. He also revealed that high frequency sound destroys the yeast, improves the appearance and inhibits the granulation of the product. These observations are very important, because crystallization was and still is one of the most serious problems that appear during the processing and marketing of honey.

The innovative idea of liquefying honey with the use of a high frequency sound was abandoned as expensive, since liquefaction of honey at that frequency demands a relatively longer time period (30 min). Later, Liebl (1977) decreased the time of honey liquefaction to 30 s using ultrasonic waves at a higher frequency (18 kHz). He was able to liquefy 1 500 kg of honey in 1 h, which reduced the cost significantly.

For unknown reasons the use of ultrasonic waves in honey processing has not been promoted as it should have been. There are as yet no studies on the effects of these waves on the quality of the honey, the changes in its chemical characteristics or the period that honey will remain in the liquid state after treatment.

In this work, results on the effects of ultrasonic waves on some chemical characteristics of honey are presented. Moreover, the time of recrystallization of the samples after liquefaction with ultrasonic waves is given.

## MATERIALS AND METHODS

Ten 300 g samples of crystallized honey were divided into 3 equal parts and were subjected to the following treatments: a) liquefaction using a MSE Soniprep 150 Ultrasonic Disintegrator, equipped with a titanium probe designated to operate at a nominal frequency of 23 kHz; b) liquefaction by heating at 60°C for 30 min in a water bath (the thermomether was in the water); or c) no treatment at all (control).

The samples were analyzed for hydroxymethylfurfural (HMF) by the Winkler method (1955) and for the diastase activity with the Schade *et al* method (1958). In addition moisture, pH and electrical conductivity were estimated as recommended by FAO/WHO (1970).

After liquefaction and analysis, the samples were stored at room temperature (25 ± 4°C) in sealed vials and the time of their recrystallization was recorded.

Five of the samples were blossom honey (numbers 1, 4, 6, 8, and 9) with no predominant pollen grains. The rest were honeydew honey from pine forest (numbers 2, 3, 5, 7 and 10).

Statistical analysis was performed using factorial ANOVA and paired *t*-test. Differences were assessed by the least significant difference (LSD) test.

## RESULTS AND DISCUSSION

The time that was needed for the liquefaction of honey by ultrasonic waves was not the same for all the samples but fluctuated from 18 to 25 mins, though the coefficient of variability (CV%) was small (9.4%) (table I). Differences could be due to the original granulated condition of samples which impeded the direct immersion of the probe tip of the instrument into the honey to an optimum depth. The differences could also be due to the nature of samples and other undefined factors. The energy also varied according to the time of treatment from 0.1056 to 0.1466 kWh. The temperature during sonication was not stable but gradually increased to the maximum shown in table I.

The combined effect of temperature 78.2 ± 1.03°C ($\bar{x}$ ± se), time 22.3 ± 0.67 mins and energy of high frequency vibrations 0.1308 ± 0.004 kWh, results in an increase of HMF, calculated at 85.7% of the untreated

**Table I.** Some technical parameters of honey liquefied by ultrasonic waves (23 kHz).

| *Sample number* | *Time (mins)* | *Temperature (°C)* | *Energy (kWh)* |
|---|---|---|---|
| 1 | 24 | 79 | 0.1408 |
| 2 | 21 | 76 | 0.1232 |
| 3 | 22 | 76 | 0.1290 |
| 4 | 18 | 76 | 0.1056 |
| 5 | 24 | 79 | 0.1408 |
| 6 | 25 | 82 | 0.1466 |
| 7 | 21 | 80 | 0.1232 |
| 8 | 21 | 76 | 0.1232 |
| 9 | 24 | 80 | 0.1408 |
| 10 | 23 | 78 | 0.1348 |
| Means | 22.3 | 78.2 | 0.1308 |
| SD [a] | 2.1 | 2.2 | 0.0123 |
| CV% | 9.4 | 2.8 | 9.4 |

[a] SD: standard deviation.

**Table II.** The effect of ultrasonic waves (23 kHz) and heat treatment (60°C for 30 min) on the HMF levels.

| *Sample number* | *Control (ppm)* | *Ultrasonic treatment* | | *Heat treatment* | |
|---|---|---|---|---|---|
| | | *ppm* | *Increase (%)* | *ppm* | *Increase (%)* |
| 1 | 8.0 | 16.1 | 101.2 | 22.6 | 182.5 |
| 2 | 12.2 | 15.0 | 22.9 | 16.7 | 36.8 |
| 3 | 5.8 | 6.9 | 18.9 | 13.2 | 127.5 |
| 4 | 8.7 | 15.0 | 72.4 | 17.3 | 98.8 |
| 5 | 4.3 | 9.3 | 116.2 | 8.2 | 90.6 |
| 6 | 5.1 | 10.4 | 103.9 | 14.7 | 188.2 |
| 7 | 2.8 | 6.9 | 146.4 | 8.2 | 192.8 |
| 8 | 4.6 | 8.0 | 73.9 | 12.4 | 169.5 |
| 9 | 7.0 | 12.0 | 71.4 | 11.4 | 62.8 |
| 10 | 6.4 | 14.7 | 129.6 | 15.2 | 137.5 |
| Means [a] | 6.5 [c] | 11.4 [d] | 85.7 | 14.0 [e] | 128.7 |
| SD [b] | | | 42.2 | | 55.2 |
| CV% | | | 49.2 | | 42.9 |

[a] Means with different letters are significantly different from each other according to LSD test; *F* value = 8.295; LSD value = 2.511 at $p = 0.010$. [b] SD: standard deviation.

samples (table II). The HMF of samples treated by heat increased to 128.7% compared with controls. The differences among averages of sonicated samples (11.4 ± 1.4 ppm) and heated ones (14.0 ± 1.4 ppm), were significant ($p < 0.010$). Thus, under the strict experimental conditions of this study, ultrasonic treatment excels heating by giving a smaller increase in HMF.

Ultrasonic energy negatively affects the diastase activity of samples. As indicated in table III, the averages of samples that received no treatment (20.8 ± 1.8 DN), and those that were treated by ultrasound (17.4 ± 1.5 DN) or heating (15.9 ± 1.2 DN) are significantly different ($p < 0.010$). In contrast to the HMF results, the difference between averages of ultrasonic and heat treatment was not significant ($p > 0.010$). The average decrease of diastase activity after ultrasonic treatment was 16.2% and after heat 23.1%. The large CV% indicates that factors other than sonication or heat may affect diastase activity. This may also indicate different behavior of the samples.

Table IV shows the effect of ultrasonic waves and heat on the moisture content, electrical conductivity and pH of honey. None of these chemical parameters was affected significantly by the treatments.

The samples that were liquefied by the ultrasonic procedure remained in the liquified state for 344.3 ± 38.8 days and after heat treatment for 282.0 ± 85.6 days (table V). The differences were not significant ($p > 0.05$, *t*-test = 1.257).

From the above results it can be concluded that liquefaction of honey with ultrasonic waves at 23 kHz may have a negative effect upon the quality of the product. Comparing this procedure with that of heat we found smaller increase of HMF. The lesser reduction in diastase activity and the

**Table III.** The effect of ultrasonic waves (23 kHz) and heat treatment (60°C for 30 min) on the diastase activity of honey.

| *Sample number* | *Control DN* | *Ultrasonic treatment* | | *Heat treatment* | |
|---|---|---|---|---|---|
| | | *DN* | *Decrease (%)* | *DN* | *Decrease (%)* |
| 1 | 15.2 | 12.1 | 20.3 | 10.3 | 32.2 |
| 2 | 19.1 | 14.8 | 22.5 | 16.8 | 12.0 |
| 3 | 25.7 | 19.0 | 26.0 | 17.1 | 33.4 |
| 4 | 17.0 | 15.7 | 7.6 | 14.8 | 12.9 |
| 5 | 32.6 | 26.4 | 19.0 | 21.7 | 33.4 |
| 6 | 17.6 | 16.5 | 6.2 | 13.3 | 24.4 |
| 7 | 24.5 | 20.3 | 17.1 | 21.6 | 11.8 |
| 8 | 18.9 | 14.6 | 22.7 | 14.5 | 23.2 |
| 9 | 14.0 | 12.0 | 14.2 | 11.0 | 21.4 |
| 10 | 24.3 | 22.7 | 6.5 | 17.9 | 26.3 |
| Means [a] | 20.8 [c] | 17.4 [d] | 16.2 | 15.9 [d] | 23.1 |
| SD [b] | | | 7.2 | | 8.6 |
| CV% | | | 44.8 | | 37.2 |

[a] Means with different letters are significantly different from each other according to LSD test; *F* value = 22.12; LSD value = 2.195 at $p = 0.010$. [b] SD: standard deviation.

**Table IV.** The effect of ultrasonic waves (23 kHz) and heat treatment (60°C for 30 min) on the moisture, electrical conductivity of honey.

| *Sample number* | *Moisture (%)* | | | *Electrical conductivity* | | | *pH* | | |
|---|---|---|---|---|---|---|---|---|---|
| | *Control* | *Ultrasound* | *Heat* | *Control* | *Ultrasound* | *Heat* | *Control* | *Ultrasound* | *Heat* |
| 1 | 20.1 | 18.2 | 17.8 | 3.0 | 2.8 | 3.0 | 3.5 | 3.8 | 3.8 |
| 2 | 18.2 | 17.5 | 17.5 | 9.3 | 9.0 | 9.8 | 3.7 | 3.8 | 3.8 |
| 3 | 17.4 | 16.3 | 17.5 | 11.8 | 11.0 | 12.0 | 4.8 | 4.6 | 4.8 |
| 4 | 16.4 | 16.3 | 16.5 | 4.1 | 4.0 | 3.8 | 3.6 | 3.7 | 3.6 |
| 5 | 19.0 | 18.4 | 19.0 | 14.3 | 13.8 | 15.2 | 4.6 | 4.8 | 4.7 |
| 6 | 18.0 | 17.1 | 17.8 | 3.6 | 3.4 | 3.5 | 3.5 | 3.4 | 3.6 |
| 7 | 16.5 | 16.4 | 16.6 | 9.0 | 8.4 | 9.5 | 4.5 | 4.7 | 4.6 |
| 8 | 18.2 | 17.4 | 16.9 | 3.0 | 3.9 | 3.7 | 4.2 | 4.0 | 4.3 |
| 9 | 17.4 | 17.5 | 17.2 | 3.9 | 4.0 | 3.9 | 3.5 | 3.4 | 3.7 |
| 10 | 16.8 | 16.3 | 15.4 | 12.0 | 11.0 | 12.4 | 3.9 | 3.8 | 3.1 |
| Means [a] | 17.8 [c] | 17.1 [c] | 17.2 [c] | 7.4 [c] | 7.1 [c] | 7.7 [c] | 3.9 [c] | 4.0 [c] | 4.0 [c] |
| SD [b] | 1.15 | 0.79 | 1.00 | 4.35 | 3.97 | 4.59 | 0.50 | 0.51 | 0.32 |
| CV% | 6.4 | 4.6 | 5.8 | 58.7 | 55.9 | 59.6 | 12.8 | 12.7 | 8.0 |

[a] Means with the same letter are not significant different from each other ($p > 0.05$). [b] SD: standard deviation.

**Table V.** Crystallization of honeys that were liquefied by ultrasonic waves and heat treatment.

| *Sample number* | *Days after treatment* | |
|---|---|---|
| | *Ultrasound* | *Heat* |
| 1 | 177 | 177 |
| 2 | 452 | 288 |
| 3 | 465 | 288 |
| 4 | 183 | 190 |
| 5 | 460 | 288 |
| 6 | 265 | 202 |
| 7 | 450 | 450 |
| 8 | 276 | 234 |
| 9 | 259 | 253 |
| 10 | 456 | 450 |
| Means [a] | 344.3 [c] | 282.0 [c] |
| se [b] | 38.8 | 85.6 |

[a] Means with the same letter are not significantly different from each other according to the *t*-test; *T* statistic = 1.257; Sig level = 0.224; p = 0.05. [b] se: standard error.

longer duration of the liquefied state of sonicated samples, cannot be invoked since the differences were not significant. We encourage the continuation of efforts for the most effective use of ultrasonic procedures. An effort should be made to reduce the ultrasonic disadvantage of the increase in temperature by surrounding the samples with a cooling jacket through which a suitable cooling liquid *(eg,* iced water or alcohol) can be circulated. Furthermore, different frequencies and different effects upon monofloral honeys should be examined.

**Résumé — Liquéfaction par les ultrasons du miel cristallisé.** On sait d'après la bibliographie (Kalogereas, 1955 ; Liebl, 1977) que le miel cristallisé peut être liquéfié par les ultrasons. Le but de ce travail est d'étudier l'action des ultrasons sur quelques caractéristiques du miel, condition importante pour leur utilisation en technologie du miel. Il a été réalisé à l'Institut d'apiculture de l'université Aristote de Thessalonique. Dix échantillons de miel cristallisés (5 de miel toutes fleurs et 5 de miellat de pin) ont été répartis en 3 groupes contenant 300 g de chacun des échantillons. Le premier groupe a été liquéfié aux ultrasons à 23 kHz à l'aide d'un désintégrateur à ultrasons MSE Soniprep 150. Le second groupe a été liquéfié par la chaleur (60°C pendant 30 min) et le 3$^{e}$ groupe a servi de témoin. Les caractéristiques suivantes ont été notées : teneur en HMF, activité diastasique, conductibilité électrique, pH, teneur en eau. On a en outre enregistré le temps de recristallisation à température ambiante (25 ± 4°C). Le tableau I donne les caractéristiques des miels liquéfiés aux ultrasons. Il a fallu en moyenne 22,3 ± 0,67 min pour liquéfier les échantillons. Pendant ce temps la température est montée régulièrement et la température maximum atteinte est indiquée dans le tableau I (moyenne : 78,2 ± 1,3°C). L'énergie nécessaire à la liquéfaction dépend de la durée et a atteint en moyenne 0,1308 ± 0,004 kWh. La durée d'application des ultrasons et la température influencent de façon négative la qualité du miel. La teneur en HMF est montée de 6,5 ± 0,8 ppm (témoin) à 11,4 ± 1,4 ppm, soit une augmentation de 81,1% (tableau II). L'activité diastasique a été réduite de 20,8 ± 1,8 DN à 17,4 ± 1,5 DN, soit une baisse de 16,2% (tableau III). Les différences entre le témoin et les miels traités aux ultrasons étaient significatives ($p < 0,01$). Si l'on compare le traitement aux ultrasons et le traitement thermique, on peut dire que la teneur en HMF est moins élevée par les ultrasons que par la température ($p < 0,01$). Il n'y a en revanche pas de différence significative en ce qui concerne l'activité diastasique ($p > 0,01$). La conductibilité électrique, le pH et la teneur en eau n'ont été que très peu modifiés par les 2 traitements (tableau IV). Le temps de recristallisation a été plus long pour les échantillons traités aux ultrasons

(344,3 ± 38,8 j) que pour ceux liquéfiés par la chaleur (282,0 ± 85,6 j). Mais cette différence n'est pas statistiquement significative ($p > 0,05$).

**cristallisation / miel / ultrason / HMF / activité diastasique**

**Zusammenfassung — Verflüssigung kristallisierten Honigs durch Ultraschall-Wellen.** Aus früheren Forschungsarbeiten (Kalogereas, 1955; Liebl, 1977) geht hervor, dass kristallisierter Honig mit Ultraschall verflüssigt werden kann. Ziel dieser Arbeit war es, den Einfluss des Ultraschalls auf einige Eigenschaften des Honigs zu untersuchen, als wichtige Voraussetzung ihrer Anwendung bei der Honigverarbeitung. Die Arbeit wurde am Institut der Aristoteles-Universität in Thessaloniki durchgeführt. Zehn Proben (5 Blütenhonige und 5 Kiefernhonige) wurden in 3 Gruppen geteilt. Die erste Gruppe wurde mit Ultraschall bei 23 kHz mit Hilfe eines MSE Soniprep 150 Ultrasonic Disintegrator verflüssigt. Die zweite Gruppe wurde durch Erwärmen auf 60°C (für 30 Minuten) verflüssigt, und die dritte Gruppe blieb unbehandelt (Kontrolle). Jede Probe wog 300 g. Die Proben wurden auf den HMF-Wert, Diastase, elektrische Leitfähigkeit, pH Wert und Wassergehalt geprüft. Zusätzlich wurde die Zeit der Wiederkristallisation der Proben bei Raumtemperatur (25 ± 4°C) beobachtet. In Tabelle I sind einige Parameter angegeben, die bei der Verflüssigung des Honigs mit Ultraschall beobachtet wurden. Durchschnittlich wurden 22,3 ± 0,67 Minuten (x SE) für die Verflüssigung der Proben benötigt. In diesem Zeitraum stieg die Temperatur ständig und erreichte ihren höchsten Wert, der aus Tabelle I zu ersehen ist, im Durchschnitt 78,2 ± 1,3°C. Die Energie, die für die Verflüssigung benötigt wurde, hing von der Zeit ab, und betrug durchschnittlich 0,1038 ± 1,004 KWh. Die Dauer der Anwendung des Ultraschalls und die Temperatur beeinflußten die Honigqualität negativ. Der HMF-Wert stieg von 6,5 ± 0,8 ppm (Kontrolle) auf 11,4 ± 1,4 ppm, eine Erhöhung also um 81,1% (Tabelle II). Die Diastase wurde von 20,8 ± 1,8 DN auf 17,4 ± 1,5 DN reduziert, also um 16,2% (Tabelle III). Die Unterschiede zwischen dem mit Ultraschall behandelten Honig und der Kontrolle waren signifikant ($P < 0,010$). Die Auswirkungen der Ultraschall-Behandlung und der Wärme-Behandlung auf die Honigqualität vergleichend, wurde festgestellt, dass Ultraschall den HMF-Wert weniger erhöht als die Temperatur ($P < 0,010$). Die Unterschiede der Diastase-Aktivität waren statistisch nicht signifikant ($P > 0,010$). Die elektrische Leitfähigkeit, der pH Wert und der Wassergehalt wurden durch beide Behandlungen nur geringfügig beeinflusst (Tabelle IV). Die Zeit der Wiederkristallisation war grösser bei den Proben, die mit Ultraschall verflüssigt wurden (344,3 ± 38,8 Tage) als bei denen, die durch Erwärmen verflüssigt wurden (282 ± 85,6 Tage). Dieser Unterschied aber ist statistisch nicht signifikant ($P > 0,05$).

**Honig / Kristallisierung / Ultraschall / HMF / Diastase**

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